

THE CLAIMS

1) A method of differentiating adipose stromal cells into osteoblasts, comprising: culturing said cells in a composition comprising a medium capable of supporting the growth of fibroblasts and differentiation inducing amounts of β -glycerophosphate and ascorbic acid and/or ascorbic-2 phosphate.

2) The method of claim 1, wherein said amounts are about 2-20 mM β -glycerophosphate and about 20-75 μ M ascorbic acid and/or ascorbic-2 phosphate.

10 3) The method of claim 2 wherein said amounts are about 5-15 mM β -glycerophosphate and about 40-60 μ M ascorbic acid and/or ascorbic-2 phosphate.

15 4) The method of claim 3, wherein said amounts are about 10 mM β -glycerophosphate and about 50 μ M ascorbic acid and/or ascorbic-2 phosphate.

5) The method of claim 1, wherein said medium is selected from the group consisting of: DMEM, α MEM and BME.

20 6) The method of claim 1, wherein said medium further comprises about 5-20% fetal calf serum.

7) The method of claim 1, wherein said medium further comprises one or more bone morphogenetic proteins.

25 8) The method of claim 1 wherein said cells are mammalian.

9) The method of claim 8 wherein said cells are human.

30 10) A method of identifying compounds that affect osteoblast differentiation,

comprising:

a) culturing adipose stromal cells in the presence and absence of a compound to be tested for effect on osteoblast differentiation in a composition which comprises a medium capable of supporting the growth of fibroblasts and differentiation inducing amounts of β -glycerophosphate and ascorbic acid and/or ascorbic-2 phosphate; and

b) comparing osteoblast differentiation in said cells cultured in the presence of said compound to that of said cells cultured in the absence of said compound.

10 11) The method of claim 10, wherein said amounts are about 2-20 mM β - glycerophosphate and about 20-75 μ M ascorbic acid and/or ascorbic-2 phosphate.

15 12) The method of claim 11, wherein said amounts are about 5-15 mM β - glycerophosphate and about 40-60 μ M ascorbic acid and/or ascorbic-2 phosphate.

13) The method of claim 12, wherein said amounts are about 10 mM β - glycerophosphate and about 50 μ M ascorbic acid and/or ascorbic-2 phosphate.

20 14) A method of improving a subject's bone structure, comprising:

a) culturing adipose stromal cells in a composition which comprises a medium capable of supporting the growth of fibroblasts and differentiation inducing amounts of β -glycerophosphate and ascorbic acid and/or ascorbic-2 phosphate; and
b) introducing said osteoblasts into a surgery or fracture site of said subject.

25 15) The method of claim 14, wherein said amounts are about 2-20 mM β - glycerophosphate and about 20-75 μ M ascorbic acid and/or ascorbic-2 phosphate.

30 16) The method of claim 15, wherein said amounts are about 5-15 mM β - glycerophosphate and about 40-60 μ M ascorbic acid and/or ascorbic-2 phosphate.

17) The method of claim 16, wherein said amounts are about 10 mM β -glycerophosphate and about 50 μ M ascorbic acid and/or ascorbic-2 phosphate.

18) The method of claim 14, wherein said adipose stromal cells are isolated
5 from said subject.

19) The method of claim 14, wherein said medium is selected from the group consisting of: DMEM, α MEM and BME.

10 20) The method of claim 14, wherein said medium further comprises about 5-
20% fetal calf serum.

15 21) The method of claim 14, wherein said medium further comprises one or
more bone morphogenetic proteins.

22) The method of claim 14, wherein said subject is mammalian.

23) The method of claim 22, wherein said subject is human.

20 24) The method of claim 14, wherein said osteoblasts are introduced in
admixture with a composition useful in the repair of bone wounds, bone defects and/or
bone disorders.

25 25) The method of claim 14, wherein a nucleotide sequence of interest is
introduced into said adipose stromal cells or said osteoblasts.

30 26) A composition comprising adipose stromal cells, a medium capable of
supporting the growth of fibroblasts and amounts of β -glycerophosphate and ascorbic
acid and/or ascorbic-2 phosphate sufficient to induce the differentiation of said stromal
cells into osteoblasts.

27) The composition of claim 26, wherein said amounts are about 2-20 mM β -glycerophosphate and about 20-75 μ M ascorbic acid and/or ascorbic-2 phosphate.

5 28) The composition of claim 27, wherein said amounts are about 5-15 mM β -glycerophosphate and about 40-60 μ M ascorbic acid and/or ascorbic-2 phosphate.

29) The composition of claim 28, wherein said amounts are about 10 mM β -glycerophosphate and about 50 μ M ascorbic acid and/or ascorbic-2 phosphate.

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30) The composition of claim 26, wherein said stromal cells are human.

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